L1 ANSWER 1 OF 10 MEDLINE on STN
AN 2006429889 IN-PROCESS
DN PubMed ID: 16851563
TI Conformational distribution of a 14-residue peptide in solution: a fluorescence resonance energy transfer study.
AU Tucker Matthew J; Oyola Rolando; Gai Feng
CS Department of Chemistry, University of Pennsylvania, Philadelphia, Pennsylvania 19104, USA.
NC GM-065978 (NIGMS)
RR-13456 (NCRR)
SO The journal of physical chemistry. B, Condensed matter, materials, surfaces, interfaces & biophysical, (2005 Mar 17) Vol. 109, No. 10, pp. 4788-95.
Journal code: 101157530. ISSN: 1520-6106.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
(RESEARCH SUPPORT, N.I.H., EXTRAMURAL)
(RESEARCH SUPPORT, U.S. GOV'T, NON-P.H.S.)
LA English
FS NONMEDLINE; IN-PROCESS; NONINDEXED; Priority Journals
ED Entered STN: 21 Jul 2006
Last Updated on STN: 14 Dec 2006
AB We demonstrate here that a nitrile-derivatized phenylalanine residue, p-cyanophenylalanine (Phe(CN)), and tryptophan (Trp) constitute a novel donor-acceptor pair for fluorescence resonance energy transfer (FRET). The Forster distance of this ***FRET pair*** was determined to be approximately 16 A and hence is well suited for determining relatively short separation distances. To validate the applicability of this ***FRET pair*** in conformational studies, we studied the conformational heterogeneity of a 14-residue amphipathic peptide, Mastoparan X (MPx peptide), in water and 7 M urea solution as well as at different temperatures. Specifically, seven nitrile-derivatized mutants of the MPx peptide, each containing a Phe(CN) residue that replaces different positions along the peptide sequence (i.e., from position 5 to 11) and serves as a resonance energy donor to the native Trp residue at position 3, were studied spectroscopically. The FRET efficiencies obtained from these peptides allowed us to gain a global picture regarding the conformational distribution of the MPx peptide in different environments. Our results suggest that the MPx molecules exist in water as an ensemble of rather compact conformations, with a radius of gyration of approximately 4.2 A, whereas in 7 M urea the radius of gyration increases to approximately 6.5 A, indicating that the peptide conformations become more extended under this condition. However, we found that temperature had only a negligible effect on the size of the MPx peptide, underlining the difference between the thermally and chemically denatured states of ***polypeptides***. The application of the Gaussian chain or the wormlike chain model allowed us to further obtain the probability distribution function of the separation distance between any two residues along the peptide sequence. We found that the effective bond length of the MPx peptide, obtained by using the Gaussian chain model, is 2.78 A in water and 4.28 A in 7 M urea.

=> s FRET (w) pair and substrate
L2 56 FRET (W) PAIR AND SUBSTRATE

=> s fluorescein and tetramethylrhodamine
L3 1192 FLUORESCEIN AND TETRAMETHYLRHODAMINE

=> l2 and l3
L2 IS NOT A RECOGNIZED COMMAND
The previous command name entered was not recognized by the system.
For a list of commands available to you in the current file, enter

"HELP COMMANDS" at an arrow prompt (=>).

=> s l2 and l3
L4 12 L2 AND L3

=> d 1-5 all

L4 ANSWER 1 OF 12 MEDLINE on STN
AN 2005658342 IN-PROCESS
DN PubMed ID: 16338388
TI A Homogeneous FRET Assay System for Multiubiquitin Chain Assembly and Disassembly.
AU Gururaja Tarikere L; Pray Todd R; Lowe Raymond; Dong Guoqiang; Huang Jianing; Daniel-Issakani Sarkiz; Payan Donald G
SO Methods in enzymology, (2005) Vol. 399, pp. 663-82.
Journal code: 0212271. ISSN: 0076-6879.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS NONMEDLINE; IN-DATA-REVIEW; IN-PROCESS; NONINDEXED; Priority Journals
ED Entered STN: 18 Dec 2005
Last Updated on STN: 13 Dec 2006
AB Ubiquitin (Ub, 76aa) is a small highly conserved protein present universally in eukaryotic cells. Covalent attachment of (Ub)(n) to target proteins is a well-known posttranslational modification that has been implicated in a wide array of cellular processes including cell biogenesis. Ubiquitin polymerization by the Ub activation-conjugation-ligation cascade and the reverse disassembly process catalyzed by Ub isopeptidases largely regulate ***substrate*** protein targeting to the 26S proteasome. Ub chains of four or more subunits attached by K48 isopeptide linkages have been shown to be necessary for the 26S proteasome association and subsequent degradation of protein molecules. To better understand this protein degradation event, it is important to develop Ub polymerization and depolymerization assays that monitor every reaction step involved in Ub attachment to, or detachment from, ***substrate*** protein molecules. In this chapter, we describe homogeneous, easy-to-use, nonradioactive, complementary continuous fluorescence assays capable of monitoring the kinetics of Ub chain formation by E3 Ub ligases, and their hydrolysis by isopeptidases, which rely on mixing a 1:1 population of fluorophore-labeled Ub molecules containing a ***FRET pair*** . The proximity of ***fluorescein*** (donor) and ***tetramethylrhodamine*** (acceptor) in Ub polymers results in ***fluorescein*** quenching on ligase-induced Ub chain assembly. Conversely, a dramatic enhancement of ***fluorescein*** emission was observed on Ub chain disassembly because of isopeptidase activity. These assays thus provide a valuable tool for monitoring Ub ligase and isopeptidase activities using authentic Ub monomers and polymers as ***substrates***. Screening of a large number of small molecule compound libraries in a high-throughput fashion is achievable, warranting further optimization of these assays.

L4 ANSWER 2 OF 12 MEDLINE on STN
AN 2003533298 MEDLINE
DN PubMed ID: 14611757
TI Development of a novel FRET immunosensor technique.
AU Lichlyter Darcy J; Grant Sheila A; Soykan Orhan
CS Department of Biological Engineering, University of Missouri-Columbia, 250 Ag. Engineering Building, Columbia, MO 65211, USA.
SO Biosensors & bioelectronics, (2003 Nov 30) Vol. 19, No. 3, pp. 219-26.
Journal code: 9001289. ISSN: 0956-5663.
CY England: United Kingdom
DT (EVALUATION STUDIES)
Journal; Article; (JOURNAL ARTICLE)
(VALIDATION STUDIES)

LA English
FS Priority Journals
EM 200407
ED Entered STN: 13 Nov 2003
Last Updated on STN: 28 Jul 2004
Entered Medline: 27 Jul 2004

AB We report on a novel technique to develop an optical immunosensor based on fluorescence resonance energy transfer (FRET). IgG antibodies were labeled with acceptor fluorophores while one of three carrier molecules (protein A, protein G, or F(ab')2 fragment) was labeled with donor fluorophores. The carrier molecule was incubated with the antibody to allow specific binding to the Fc portion. The labeled antibody-protein complex was then exposed to specific and nonspecific antigens, and experiments were designed to determine the 'in solution' response. The paper reports the results of three different donor-acceptor ***FRET pairs***, ***fluorescein*** isothiocyanate/ ***tetramethylrhodamine*** isothiocyanate, Texas Red/Cy5, and Alexa Fluor 546/Alexa Fluor 594. The effects of the fluorophore to protein conjugation ratio (F/P ratio) and acceptor to donor fluorophore ratios between the antibody and protein (A/D ratio) were examined. In the presence of specific antigens, the antibodies underwent a conformational change, resulting in an energy transfer from the donor to the acceptor fluorophore as measured by a change in fluorescence. The non-specific antigens elicited little or no changes. The Alexa Fluor ***FRET pair*** demonstrated the largest change in fluorescence, resulting in a 35% change. The F/P and A/D ratio will affect the efficiency of energy transfer, but there exists a suitable range of A/D and F/P ratios for the ***FRET pairs***. The feasibility of the FRET immunosensor technique was established; however, it will be necessary to immobilize the complexes onto optical ***substrates*** so that consistent trends can be obtained that would allow calibration plots.

CT Antigen-Antibody Complex: AN, analysis
Antigen-Antibody Complex: CH, chemistry
Antigen-Antibody Complex: IM, immunology
*Antigens: AN, analysis
*Antigens: CH, chemistry
Antigens: IM, immunology
Biosensing Techniques: IS, instrumentation
*Biosensing Techniques: MT, methods
Comparative Study
Equipment Design
Equipment Failure Analysis
Fluorescence Polarization Immunoassay: IS, instrumentation
*Fluorescence Polarization Immunoassay: MT, methods
Fluorescence Resonance Energy Transfer: IS, instrumentation
*Fluorescence Resonance Energy Transfer: MT, methods
Immunoglobulin Fab Fragments: AN, analysis
Immunoglobulin Fab Fragments: CH, chemistry
Immunoglobulin Fab Fragments: IM, immunology
*Immunoglobulin G: AN, analysis
*Immunoglobulin G: CH, chemistry
Nerve Tissue Proteins: AN, analysis
Nerve Tissue Proteins: CH, chemistry
Nerve Tissue Proteins: IM, immunology
Reproducibility of Results
Research Support, Non-U.S. Gov't
Sensitivity and Specificity
Staphylococcal Protein A: AN, analysis
Staphylococcal Protein A: CH, chemistry
Staphylococcal Protein A: IM, immunology

CN 0 (Antigen-Antibody Complex); 0 (Antigens); 0 (G-***substrate***); 0 (Immunoglobulin Fab Fragments); 0 (Immunoglobulin G); 0 (Nerve Tissue Proteins); 0 (Staphylococcal Protein A)

L4 ANSWER 3 OF 12 MEDLINE on STN
AN 1999430268 MEDLINE
DN PubMed ID: 10500481
TI A fluorescent indicator for tyrosine phosphorylation-based insulin signaling pathways.
AU Sato M; Ozawa T; Yoshida T; Umezawa Y
CS Department of Chemistry, School of Science, University of Tokyo, Japan.
SO Analytical chemistry, (1999 Sep 15) Vol. 71, No. 18, pp. 3948-54. Journal code: 0370536. ISSN: 0003-2700.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199911
ED Entered STN: 11 Jan 2000
Last Updated on STN: 11 Jan 2000
Entered Medline: 2 Nov 1999
AB A fluorescent indicator for tyrosine phosphorylation-based insulin signaling is described. Upon binding of insulin to cell-surface insulin receptor, the receptor phosphorylates tyrosine residues of insulin receptor ***substrate*** 1 (IRS-1) in the cell. A fluorescent indicator was designed by using synthetic phosphopeptide pY939 derived from the tyrosine phosphorylation domain of IRS-1 and its target protein SH2N containing an N-terminal SH2 domain of PI 3-kinase. The SH2N protein and pY939 phosphopeptide were labeled with ***fluorescein*** (F-SH2N) and ***tetramethylrhodamine*** (T-pY939), respectively. Formation of a F-SH2N-T-pY939 complex (termed a fluorescence resonance energy-transfer (***FRET***) ***pair***) was evaluated from a change in a fluorescence emission spectrum based on FRET between the two fluorophores. The ***FRET pair*** was formed to dissociate in competition with the unlabeled pY939 phosphopeptide, resulting in a decrease in a pY939 phosphopeptide-dependent FRET emission at 580 nm and causing an increase in emission at 520 nm. Tyrosine phosphorylation by the partially purified insulin receptor of ***substrate*** peptide Y939 was detected with this formed ***FRET pair***, and resulting changes in fluorescence emission spectra were observed for insulin concentration from about 1.0 x 10(-9) to 1.0 x 10(-6) M. These results indicated that the ***FRET pair*** served as a competitive fluorescent indicator for tyrosine phosphorylation-based insulin signaling.
CT 1-Phosphatidylinositol 3-Kinase: ME, metabolism
Amino Acid Sequence
Animals
Comparative Study
Cricetinae
Fluorescein: CH, chemistry
Fluorescein: ME, metabolism
*Fluorescent Dyes: CH, chemistry
Insulin: ME, metabolism
*Insulin: PH, physiology
*Phosphopeptides: CH, chemistry
*Phosphoproteins: CH, chemistry
Phosphoproteins: ME, metabolism
Phosphorylation
Receptor, Insulin: ME, metabolism
Research Support, Non-U.S. Gov't
*Signal Transduction: PH, physiology
Spectrometry, Fluorescence: MT, methods
*Tyrosine: ME, metabolism
src Homology Domains
RN 11061-68-0 (Insulin); ***2321-07-5 (Fluorescein)***; 55520-40-6 (Tyrosine)
CN 0 (Fluorescent Dyes); 0 (Phosphopeptides); 0 (Phosphoproteins); 0 (insulin receptor ***substrate***-1 protein); 0 (phospho-Y939 peptide); EC 2.7.1.112 (Receptor, Insulin); EC 2.7.1.137 (1-Phosphatidylinositol

3-Kinase)

L4 ANSWER 4 OF 12 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
AN 2006:163943 BIOSIS
DN PREV200600178304
TI A homogeneous FRET assay system for multiubiquitin chain assembly and disassembly.
AU Gururaja, Tarikere L. [Reprint Author]; Pray, Todd R.; Lowe, Raymond; Dong, Guoqiang; Huang, Jianing; Daniel-Issakani, Sarkiz; Payan, Donald G.
CS Rigel Pharmaceut Inc, San Francisco, CA USA
SO Deshaies, RJ [Editor]. Methods Enzymol., (2005) pp. 663-682. Methods in Enzymology.
Publisher: ELSEVIER ACADEMIC PRESS INC, 525 B STREET, SUITE 1900, SAN DIEGO, CA 92101-4495 USA. Series: METHODS IN ENZYMOLOGY.
CODEN: MENZAU. ISSN: 0076-6879. ISBN: 0-12-182804-2(H).
DT Book; (Book Chapter)
LA English
ED Entered STN: 9 Mar 2006
Last Updated on STN: 9 Mar 2006
AB Ubiquitin (Ub, 76aa) is a small highly conserved protein present universally in eukaryotic cells. Covalent attachment of (Ub)(n) to target proteins is a well-known posttranslational modification that has been implicated in a wide array of cellular processes including cell biogenesis. Ubiquitin polymerization by the Ub activation-conjugation-ligation cascade and the reverse disassembly process catalyzed by Ub isopeptidases largely regulate ***substrate*** protein targeting to the 26S proteasome. Ub chains of four or more subunits attached by K48 isopeptide linkages have been shown to be necessary for the 26S proteasome association and subsequent degradation of protein molecules. To better understand this protein degradation event, it is important to develop Ub polymerization and depolymerization assays that monitor every reaction step involved in Ub attachment to, or detachment from, ***substrate*** protein molecules. In this chapter, we describe homogeneous, easy-to-use, nonradioactive, complementary continuous fluorescence assays capable of monitoring the kinetics of Ub chain formation by E3 Ub ligases, and their hydrolysis by isopeptidases, which rely on mixing a 1:1 population of fluorophore-labeled Ub molecules containing a ***FRET pair*** . The proximity of ***fluorescein*** (donor) and ***tetramethylrhodamine*** (acceptor) in Ub polymers results in ***fluorescein*** quenching on ligase-induced Ub chain assembly. Conversely, a dramatic enhancement of ***fluorescein*** emission was observed on Ub chain disassembly because of isopeptidase activity. These assays thus provide a valuable tool for monitoring Ub ligase and isopeptidase activities using authentic Ub monomers and polymers as ***substrates***. Screening of a large number of small molecule compound libraries in a high-throughput fashion is achievable, warranting further optimization of these assays.
CC Cytology - General 02502
Biochemistry studies - General 10060
Biochemistry studies - Proteins, peptides and amino acids 10064
Enzymes - General and comparative studies: coenzymes 10802
IT Major Concepts
Methods and Techniques; Enzymology (Biochemistry and Molecular Biophysics); Cell Biology
IT Chemicals & Biochemicals
ubiquitin; ***fluorescein***; E3 ubiquitin ligase [EC 6.3.2.19]; fluorophore; 26S proteasome; ***tetramethylrhodamine***; isopeptidase [EC 3.1.2.15]; multiubiquitin binding protein
IT Methods & Equipment
fluorescence resonance energy transfer imaging [FRET imaging]: laboratory techniques, imaging and microscopy techniques
IT Miscellaneous Descriptors
cellular process
ORGN Classifier

Organisms 00500
Super Taxa
Organisms
Organism Name
eukaryote (common)
Taxa Notes
Organisms
RN 60267-61-0 (ubiquitin)
2321-07-5 (***fluorescein***)
74812-49-0 (E3 ubiquitin ligase)
74812-49-0 (EC 6.3.2.19)
140879-24-9 (26S proteasome)
70281-37-7 (***tetramethylrhodamine***)
353301-20-9 (isopeptidase)
353301-20-9 (EC 3.1.2.15)

L4 ANSWER 5 OF 12 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
AN 2004:54710 BIOSIS
DN PREV200400057974
TI Development of a novel FRET immunosensor technique.
AU Lichlyter, Darcy J.; Grant, Sheila A. [Reprint Author]; Soykan, Orhan
CS Department of Biological Engineering, University of Missouri-Columbia, 250 Ag. Engineering Building, Columbia, MO, 65211, USA
grantsa@missouri.edu
SO Biosensors & Bioelectronics, (30 November 2003) Vol. 19, No. 3, pp. 219-226. print.
CODEN: BBIOE4. ISSN: 0956-5663.
DT Article
LA English
ED Entered STN: 21 Jan 2004
Last Updated on STN: 21 Jan 2004
AB We report on a novel technique to develop an optical immunosensor based on fluorescence resonance energy transfer (FRET). IgG antibodies were labeled with acceptor fluorophores while one of three carrier molecules (protein A, protein G, or F(ab')2 fragment) was labeled with donor fluorophores. The carrier molecule was incubated with the antibody to allow specific binding to the Fc portion. The labeled antibody-protein complex was then exposed to specific and nonspecific antigens, and experiments were designed to determine the 'in solution' response. The paper reports the results of three different donor-acceptor ***FRET pairs***, ***fluorescein*** isothiocyanate/ ***tetramethylrhodamine*** isothiocyanate, Texas Red/Cy5, and Alexa Fluor 546/Alexa Fluor 594. The effects of the fluorophore to protein conjugation ratio (F/P ratio) and acceptor to donor fluorophore ratios between the antibody and protein (A/D ratio) were examined. In the presence of specific antigens, the antibodies underwent a conformational change, resulting in an energy transfer from the donor to the acceptor fluorophore as measured by a change in fluorescence. The non-specific antigens elicited little or no changes. The Alexa Fluor ***FRET pair*** demonstrated the largest change in fluorescence, resulting in a 35% change. The F/P and A/D ratio will affect the efficiency of energy transfer, but there exists a suitable range of AID and F/P ratios for the ***FRET pairs***. The feasibility of the FRET immunosensor technique was established; however, it will be necessary to immobilize the complexes onto optical ***substrates*** so that consistent trends can be obtained that would allow calibration plots.
CC Biochemistry studies - General 10060
Biochemistry studies - Proteins, peptides and amino acids 10064
Immunology - General and methods 34502
IT Major Concepts
Biochemistry and Molecular Biophysics; Equipment Apparatus Devices and Instrumentation; Methods and Techniques
IT Chemicals & Biochemicals
Alexa Fluor 546/Alexa Fluor 594: donor-acceptor ***FRET***

pair; F(ab')-2 fragment: carrier molecule; IgG antibodies [immunoglobulin G antibodies]; Texas Red/Cy5: donor-acceptor ***FRET pair***; acceptor fluorophores; donor fluorophores; ***fluorescein*** isothiocyanate/***tetramethylrhodamine*** isothiocyanate: donor-acceptor ***FRET pair***; protein A: carrier molecule; protein G: carrier molecule

IT Methods & Equipment

fluorescence resonance energy transfer immunosensor technique [FRET immunosensor technique]: immunologic techniques, laboratory techniques, spectrum analysis techniques; optical immunosensor: laboratory equipment

IT Miscellaneous Descriptors

acceptor/donor fluorophore ratios; fluorophore/protein conjugation ratio; technical development

RN 521-18-6 (protein A)

=> s IAEDANS and fluorescein

L5 130 IAEDANS AND FLUORESCEIN

```
=> s FRET(w) pair and substrate
L1          56 FRET(W) PAIR AND SUBSTRATE

=> s IAEDANS and fluorescein
L2         130 IAEDANS AND FLUORESCEIN

=> s fluorescein and tetramethylrhodamine
L3        1192 FLUORESCEIN AND TETRAMETHYLRHODAMINE

=> s EDANS and DABCYL
L4         193 EDANS AND DABCYL

=> s BODIPY fluorescein
L5          14 BODIPY FLUORESCEIN

=> s phydoerythrin and Cy5
L6           0 PHYDOERYTHRIN AND CY5

=> s phycoerythrin and Cy5
L7         170 PHYCOERYTHRIN AND CY5

=> s pyrene and coumarin
L8         770 PYRENE AND COUMARIN

=> s l1 and l2
L9           1 L1 AND L2

=> d l9 all

L9   ANSWER 1 OF 1  CAPLUS  COPYRIGHT 2006 ACS on STN
AN   2006:170631  CAPLUS
DN   144:249410
ED   Entered STN:  24 Feb 2006
TI   FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing
     semisynthetic dual-labeled proximity sensor peptide-containing Crk-II
     construct, and potential screening use
IN   Muir, Tom; Cotton, Graham
PA   USA
SO   U.S. Pat. Appl. Publ., 19 pp., Cont. of U.S. Ser. No. 483,543, abandoned.
     CODEN: USXXCO
DT   Patent
LA   English
INCL 435007100
CC   7-1 (Enzymes)
     Section cross-reference(s): 1, 34
FAN.CNT 1
     PATENT NO.          KIND   DATE       APPLICATION NO.         DATE
     ---------------     ----   --------   --------------------    --------
PI   US 2006040319        A1    20060223   US 2004-784721          20040223
PRAI US 2000-483543       B1    20000114
CLASS
 PATENT NO.       CLASS  PATENT FAMILY CLASSIFICATION CODES
 --------------   -----  ----------------------------------------------
 US 2006040319    INCL   435007100
                  IPCI   G01N0033-53 [I,A]
                  IPCR   G01N0033-53 [I,A]; G01N0033-53 [I,C]
                  NCL    435/007.100
                  ECLA   C12Q001/48B; G01N033/542
AB   Compns. and methods are provided for identifying conformational changes in
     polypeptides related to the activity or biol. state of the polypeptide.
     Semisynthetic polypeptides are prepared comprising at least two
     proximity-sensor peptides, the resultant composition capable of detectably
     indicating the activity of biol. state of the polypeptide.  Such compns.
     may be used to identify modulators of the polypeptides as well as
```

modulators of mols. which interact with the polypeptide, such as protein kinases which act on protein kinase targets. More specifically, the invention provides a biosensor for c-Abl phosphorylation of the Crk-II adapter protein. The structure of a dual-labeled, semisynthetic, recombinantly prepared composition comprising the protein kinase adapter protein
Crk-II which is capable of reporting phosphorylation by c-Abl is disclosed. Generation of Rh-(Crk-II)-Fl (Rh = tetramethylrhodamine; Fl = ***fluorescein***) by solid-phase protein ligation and phosphorylation of Rh-(Crk-II)-Fl by full-length c-Abl is reported. It was shown that Rh-(Crk-II)-Fl is a fluorescence biosensor for c-Abl phosphorylation of Crk-II utilizing FRET for c-Abl determination One potential use of this biosensor
is in the rapid screening of c-Abl kinase inhibitors.

ST fluorescence biosensor cAbl kinase detn CrkII phosphorylation FRET; proximity sensor peptide fluorescence biosensor protein kinase detn screening; ***FRET pair*** tetramethylrhodamine ***fluorescein*** proximity sensor peptide fluorescence biosensor

IT Proteins
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); SPN (Synthetic preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(CRKII, fusion products; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Mus musculus
(Crk-II of; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Dephosphorylation, biological
Drug screening
Fluorescent indicators
Phosphorylation, biological
Protein engineering
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Enzymes, analysis
RL: ANT (Analyte); ANST (Analytical study)
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein motifs
(SH2 domain; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein motifs
(SH3 domain; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Conformational transition
(activity-related, detection of; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Phosphorylation
(enzymic; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Fluorescence resonance energy transfer
(fluorescent label pair for; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein sequences
(of Crk-II construct; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT Post-translational processing
(of peptide ***substrate***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT Biosensors
(optical; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT Solid phase synthesis
(peptide, solid-phase protein ligation; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT Amino acids, uses
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(proximity sensor peptide containing fluorescent amino acid derivative; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT Oligopeptides
RL: ARG (Analytical reagent use); SPN (Synthetic preparation); ANST (Analytical study); PREP (Preparation); USES (Uses)
(proximity sensor; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT Proteins
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(recombinant, ***substrates***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT Peptides, uses
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(***substrate***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT Phycoerythrins
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(β-, fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT 146368-14-1, Cy5
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(Cy5, fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT 9031-44-1, Kinase (phosphorylating) 98037-52-6, Abelson protein tyrosine kinase 138238-67-2, c-Abl kinase
RL: ANT (Analyte); ANST (Analytical study)
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
IT 876799-69-8P 876799-70-1P
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); SPN (Synthetic

preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(amino acid sequence; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 91-64-5, Coumarin 129-00-0, Pyrene, uses 2321-07-5, ***Fluorescein*** 6268-49-1, DABCYL 36930-63-9, ***IAEDANS*** 50402-56-7, EDANS 70281-37-7, Tetramethylrhodamine 138026-71-8, BODIPY 165599-63-3, BODIPY FL
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 876656-99-4D, conjugates with ***fluorescein***
RL: RCT (Reactant); RACT (Reactant or reagent)
(preparation of dual-labeled Crk-II; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 876808-11-6
RL: PRP (Properties)
(unclaimed protein sequence; fRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 168202-46-8 302566-59-2 876760-73-5 876760-74-6 876808-08-1 876808-09-2 876808-10-5
RL: PRP (Properties)
(unclaimed sequence; fRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

=> s l1 and l3
L10 12 L1 AND L3

=> d l10 1-5 abs

L10 ANSWER 1 OF 12 MEDLINE on STN

AB Ubiquitin (Ub, 76aa) is a small highly conserved protein present universally in eukaryotic cells. Covalent attachment of (Ub)(n) to target proteins is a well-known posttranslational modification that has been implicated in a wide array of cellular processes including cell biogenesis. Ubiquitin polymerization by the Ub activation-conjugation-ligation cascade and the reverse disassembly process catalyzed by Ub isopeptidases largely regulate ***substrate*** protein targeting to the 26S proteasome. Ub chains of four or more subunits attached by K48 isopeptide linkages have been shown to be necessary for the 26S proteasome association and subsequent degradation of protein molecules. To better understand this protein degradation event, it is important to develop Ub polymerization and depolymerization assays that monitor every reaction step involved in Ub attachment to, or detachment from, ***substrate*** protein molecules. In this chapter, we describe homogeneous, easy-to-use, nonradioactive, complementary continuous fluorescence assays capable of monitoring the kinetics of Ub chain formation by E3 Ub ligases, and their hydrolysis by isopeptidases, which rely on mixing a 1:1 population of fluorophore-labeled Ub molecules containing a ***FRET pair*** . The proximity of ***fluorescein*** (donor) and ***tetramethylrhodamine*** (acceptor) in Ub polymers results in ***fluorescein*** quenching on ligase-induced Ub chain assembly. Conversely, a dramatic enhancement of ***fluorescein*** emission was observed on Ub chain disassembly because of isopeptidase activity. These assays thus provide a valuable tool for monitoring Ub ligase and isopeptidase activities using authentic Ub monomers and polymers as ***substrates***. Screening of a large number of small molecule compound libraries in a high-throughput fashion is achievable, warranting

further optimization of these assays.

L10 ANSWER 2 OF 12 MEDLINE on STN
AB We report on a novel technique to develop an optical immunosensor based on fluorescence resonance energy transfer (FRET). IgG antibodies were labeled with acceptor fluorophores while one of three carrier molecules (protein A, protein G, or F(ab')2 fragment) was labeled with donor fluorophores. The carrier molecule was incubated with the antibody to allow specific binding to the Fc portion. The labeled antibody-protein complex was then exposed to specific and nonspecific antigens, and experiments were designed to determine the 'in solution' response. The paper reports the results of three different donor-acceptor ***FRET pairs***, ***fluorescein*** isothiocyanate/ ***tetramethylrhodamine*** isothiocyanate, Texas Red/Cy5, and Alexa Fluor 546/Alexa Fluor 594. The effects of the fluorophore to protein conjugation ratio (F/P ratio) and acceptor to donor fluorophore ratios between the antibody and protein (A/D ratio) were examined. In the presence of specific antigens, the antibodies underwent a conformational change, resulting in an energy transfer from the donor to the acceptor fluorophore as measured by a change in fluorescence. The non-specific antigens elicited little or no changes. The Alexa Fluor ***FRET pair*** demonstrated the largest change in fluorescence, resulting in a 35% change. The F/P and A/D ratio will affect the efficiency of energy transfer, but there exists a suitable range of A/D and F/P ratios for the ***FRET pairs***. The feasibility of the FRET immunosensor technique was established; however, it will be necessary to immobilize the complexes onto optical ***substrates*** so that consistent trends can be obtained that would allow calibration plots.

L10 ANSWER 3 OF 12 MEDLINE on STN
AB A fluorescent indicator for tyrosine phosphorylation-based insulin signaling is described. Upon binding of insulin to cell-surface insulin receptor, the receptor phosphorylates tyrosine residues of insulin receptor ***substrate*** 1 (IRS-1) in the cell. A fluorescent indicator was designed by using synthetic phosphopeptide pY939 derived from the tyrosine phosphorylation domain of IRS-1 and its target protein SH2N containing an N-terminal SH2 domain of PI 3-kinase. The SH2N protein and pY939 phosphopeptide were labeled with ***fluorescein*** (F-SH2N) and ***tetramethylrhodamine*** (T-pY939), respectively. Formation of a F-SH2N-T-pY939 complex (termed a fluorescence resonance energy-transfer (***FRET***) ***pair***) was evaluated from a change in a fluorescence emission spectrum based on FRET between the two fluorophores. The ***FRET pair*** was formed to dissociate in competition with the unlabeled pY939 phosphopeptide, resulting in a decrease in a pY939 phosphopeptide-dependent FRET emission at 580 nm and causing an increase in emission at 520 nm. Tyrosine phosphorylation by the partially purified insulin receptor of ***substrate*** peptide Y939 was detected with this formed ***FRET pair***, and resulting changes in fluorescence emission spectra were observed for insulin concentration from about 1.0 x 10(-9) to 1.0 x 10(-6) M. These results indicated that the ***FRET pair*** served as a competitive fluorescent indicator for tyrosine phosphorylation-based insulin signaling.

L10 ANSWER 4 OF 12 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
AB Ubiquitin (Ub, 76aa) is a small highly conserved protein present universally in eukaryotic cells. Covalent attachment of (Ub)(n) to target proteins is a well-known posttranslational modification that has been implicated in a wide array of cellular processes including cell biogenesis. Ubiquitin polymerization by the Ub activation-conjugation-ligation cascade and the reverse disassembly process catalyzed by Ub isopeptidases largely regulate ***substrate*** protein targeting to the 26S proteasome. Ub chains of four or more subunits attached by K48 isopeptide linkages have been shown to be necessary for the 26S proteasome association and subsequent degradation of protein molecules. To better

understand this protein degradation event, it is important to develop Ub polymerization and depolymerization assays that monitor every reaction step involved in Ub attachment to, or detachment from, ***substrate*** protein molecules. In this chapter, we describe homogeneous, easy-to-use, nonradioactive, complementary continuous fluorescence assays capable of monitoring the kinetics of Ub chain formation by E3 Ub ligases, and their hydrolysis by isopeptidases, which rely on mixing a 1:1 population of fluorophore-labeled Ub molecules containing a ***FRET pair*** . The proximity of ***fluorescein*** (donor) and ***tetramethylrhodamine*** (acceptor) in Ub polymers results in ***fluorescein*** quenching on ligase-induced Ub chain assembly. Conversely, a dramatic enhancement of ***fluorescein*** emission was observed on Ub chain disassembly because of isopeptidase activity. These assays thus provide a valuable tool for monitoring Ub ligase and isopeptidase activities using authentic Ub monomers and polymers as ***substrates***. Screening of a large number of small molecule compound libraries in a high-throughput fashion is achievable, warranting further optimization of these assays.

L10 ANSWER 5 OF 12 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
AB We report on a novel technique to develop an optical immunosensor based on fluorescence resonance energy transfer (FRET). IgG antibodies were labeled with acceptor fluorophores while one of three carrier molecules (protein A, protein G, or F(ab')2 fragment) was labeled with donor fluorophores. The carrier molecule was incubated with the antibody to allow specific binding to the Fc portion. The labeled antibody-protein complex was then exposed to specific and nonspecific antigens, and experiments were designed to determine the 'in solution' response. The paper reports the results of three different donor-acceptor ***FRET pairs***, ***fluorescein*** isothiocyanate/ ***tetramethylrhodamine*** isothiocyanate, Texas Red/Cy5, and Alexa Fluor 546/Alexa Fluor 594. The effects of the fluorophore to protein conjugation ratio (F/P ratio) and acceptor to donor fluorophore ratios between the antibody and protein (A/D ratio) were examined. In the presence of specific antigens, the antibodies underwent a conformational change, resulting in an energy transfer from the donor to the acceptor fluorophore as measured by a change in fluorescence. The non-specific antigens elicited little or no changes. The Alexa Fluor ***FRET pair*** demonstrated the largest change in fluorescence, resulting in a 35% change. The F/P and A/D ratio will affect the efficiency of energy transfer, but there exists a suitable range of AID and F/P ratios for the ***FRET pairs***. The feasibility of the FRET immunosensor technique was established; however, it will be necessary to immobilize the complexes onto optical ***substrates*** so that consistent trends can be obtained that would allow calibration plots.

=> d l10 6-12 abs

L10 ANSWER 6 OF 12 CAPLUS COPYRIGHT 2006 ACS on STN
AB Compns. and methods are provided for identifying conformational changes in polypeptides related to the activity or biol. state of the polypeptide. Semisynthetic polypeptides are prepared comprising at least two proximity-sensor peptides, the resultant composition capable of detectably indicating the activity of biol. state of the polypeptide. Such compns. may be used to identify modulators of the polypeptides as well as modulators of mols. which interact with the polypeptide, such as protein kinases which act on protein kinase targets. More specifically, the invention provides a biosensor for c-Abl phosphorylation of the Crk-II adapter protein. The structure of a dual-labeled, semisynthetic, recombinantly prepared composition comprising the protein kinase adapter protein
Crk-II which is capable of reporting phosphorylation by c-Abl is disclosed. Generation of Rh-(Crk-II)-Fl (Rh =

tetramethylrhodamine; Fl = ***fluorescein***) by solid-phase protein ligation and phosphorylation of Rh-(Crk-II)-Fl by full-length c-Abl is reported. It was shown that Rh-(Crk-II)-Fl is a fluorescence biosensor for c-Abl phosphorylation of Crk-II utilizing FRET for c-Abl determination One potential use of this biosensor is in the rapid screening of c-Abl kinase inhibitors.

L10 ANSWER 7 OF 12 CAPLUS COPYRIGHT 2006 ACS on STN

AB We report on a novel technique to develop an optical immunosensor based on fluorescence resonance energy transfer (FRET). IgG antibodies were labeled with acceptor fluorophores while one of three carrier mols. (protein A, protein G, or F(ab')2 fragment) was labeled with donor fluorophores. The carrier mol. was incubated with the antibody to allow specific binding to the Fc portion. The labeled antibody-protein complex was then exposed to specific and nonspecific antigens, and expts. were designed to determine the in solution' response. The paper reports the results of three different donor-acceptor ***FRET pairs***, ***fluorescein*** isothiocyanate/***tetramethylrhodamine*** isothiocyanate, Texas Red/Cy5, and Alexa Fluor 546/Alexa Fluor 594. The effects of the fluorophore to protein conjugation ratio (F/P ratio) and acceptor to donor fluorophore ratios between the antibody and protein (A/D ratio) were examined In the presence of specific antigens, the antibodies underwent a conformational change, resulting in an energy transfer from the donor to the acceptor fluorophore as measured by a change in fluorescence. The non-specific antigens elicited little or no changes. The Alexa Fluor ***FRET pair*** demonstrated the largest change in fluorescence, resulting in a 35% change. The F/P and A/D ratio will affect the efficiency of energy transfer, but there exists a suitable range of A/D and F/P ratios for the ***FRET pairs***. The feasibility of the FRET immunosensor technique was established; however, it will be necessary to immobilize the complexes onto optical ***substrates*** so that consistent trends can be obtained that would allow calibration plots.

L10 ANSWER 8 OF 12 CAPLUS COPYRIGHT 2006 ACS on STN

AB A method for detecting the binding of a test compound to a probe mol. comprises providing a test compound, the test compound having a first fluorophore bound thereto, and providing a screening ***substrate***. The screening ***substrate*** comprises a solid support, a probe mol. bound to the solid support, and a second fluorophore bound to the solid support adjacent the probe mol. An advantage of the invention is that this obviates the need for binding the second fluorophore directly to the probe mol. Preferably, the second fluorophore is bound to the solid support by a flexible linker group. This enables the second fluorophore to interrogate different positions on the probe mol., which is also bound to the solid support adjacent the linker group, enhancing the ability of the method of the invention to detect pos. binding events (specific binding of the test compound to the probe mol.). The first and second fluorophores together comprise the donor and acceptor fluorophores of a fluorescence resonance energy transfer (***FRET***) ***pair***, or a "donor/acceptor pair.". The test compound is contacted to the screening ***substrate***, and the screening ***substrate*** illuminated with light at a wavelength that is absorbed by the donor fluorophore. The transfer of energy from one to the other fluorophore is then detected, with the transfer of energy indicating the binding of the test compound to the probe. ***Substrates*** useful for carrying out the foregoing methods are also disclosed.

L10 ANSWER 9 OF 12 CAPLUS COPYRIGHT 2006 ACS on STN

AB A fluorescent indicator for tyrosine phosphorylation-based insulin signaling is described. Upon binding of insulin to cell-surface insulin receptor, the receptor phosphorylates tyrosine residues of insulin receptor ***substrate*** 1 (IRS-1) in the cell. A fluorescent

indicator was designed by using synthetic phosphopeptide pY939 derived from the tyrosine phosphorylation domain of IRS-1 and its target protein SH2N containing an N-terminal SH2 domain of PI 3-kinase. The SH2N protein and pY939 phosphopeptide were labeled with ***fluorescein*** (F-SH2N) and ***tetramethylrhodamine*** (T-pY939), resp. Formation of a F-SH2N-T-pY939 complex (termed a fluorescence resonance energy-transfer (***FRET***) ***pair***) was evaluated from a change in a fluorescence emission spectrum based on FRET between the two fluorophores. The ***FRET pair*** was formed to dissociate in competition with the unlabeled pY939 phosphopeptide, resulting in a decrease in a pY939 phosphopeptide-dependent FRET emission at 580 nm and causing an increase in emission at 520 nm. Tyrosine phosphorylation by the partially purified insulin receptor of ***substrate*** peptide Y939 was detected with this formed ***FRET pair***, and resulting changes in fluorescence emission spectra were observed for insulin concentration from about 1.0 + 10-9 to 1.0 + 10-6 M. These results indicated that the ***FRET pair*** served as a competitive fluorescent indicator for tyrosine phosphorylation-based insulin signaling.

L10 ANSWER 10 OF 12 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

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L10 ANSWER 11 OF 12 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

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tetramethylrhodamine isothiocyanate, Texas Red/Cy5, and Alexa Fluor 546/Alexa Fluor 594. The effects of the fluorophore to protein conjugation ratio (F/P ratio) and acceptor to donor fluorophore ratios between the antibody and protein (A/D ratio) were examined. In the presence of specific antigens, the antibodies underwent a conformational change, resulting in an energy transfer from the donor to the acceptor fluorophore as measured by a change in fluorescence. The non-specific antigens elicited little or no changes. The Alexa Fluor ***FRET pair*** demonstrated the largest change in fluorescence, resulting in a 35% change. The F/P and A/D ratio will affect the efficiency of energy transfer, but there exists a suitable range of A/D and F/P ratios for the ***FRET pairs***. The feasibility of the FRET immunosensor technique was established; however, it will be necessary to immobilize the complexes onto optical ***substrates*** so that consistent trends can be obtained that would allow calibration plots.

L10 ANSWER 12 OF 12 EMBASE

AB A fluorescent indicator for tyrosine phosphorylation-based insulin signaling is described. Upon binding of insulin to cell-surface insulin receptor, the receptor phosphorylates tyrosine residues of insulin receptor ***substrate*** 1 (IRS-1) in the cell. A fluorescent indicator was designed by using synthetic phosphopeptide pY939 derived from the tyrosine phosphorylation domain of IRS-1 and its target protein SH2N containing an N- terminal SH2 domain of PI 3-kinase. The SH2N protein and pY939 phosphopeptide were labeled with ***fluorescein*** (F-SH2N) and ***tetramethylrhodamine*** (T-pY939), respectively. Formation of a F-SH2N-T-pY939 complex (termed a fluorescence resonance energy-transfer (***FRET***)***pair***) was evaluated from a change in a fluorescence emission spectrum based on FRET between the two fluorophores. The ***FRET pair*** was formed to dissociate in competition with the unlabeled pY939 phosphopeptide, resulting in a decrease in a pY939 phosphopeptide- dependent FRET emission at 580 nm and causing an increase in emission at 520 nm. Tyrosine phosphorylation by the partially purified insulin receptor of ***substrate*** peptide Y939 was detected with this formed ***FRET pair***, and resulting changes in fluorescence emission spectra were observed for insulin concentration from about 1.0 x 10-9 to 1.0 x 10-6 M. These results indicated that the ***FRET pair*** served as a competitive fluorescent indicator for tyrosine phosphorylation-based insulin signaling.

```
=> s l1 and l4
L11          4 L1 AND L4

=> d 1-4 abs
```

L11 ANSWER 1 OF 4 MEDLINE on STN

AB Lysostaphin (EC. 3.4.24.75) is a protein secreted by Staphylococcus simulans biovar staphylolyticus and has been shown to be active against methicillin resistant S. aureus (MRSA). The design and synthesis of three internally quenched ***substrates*** for lysostaphin based on the peptidoglycan crossbridges of S. aureus, and their use in fluorescence resonance energy transfer (FRET) assays is reported. These ***substrates*** enabled the gathering of information about the endopeptidase activity of lysostaphin and the effect that mutations have on its enzymatic ability. Significant problems with the inner filter effect and ***substrate*** aggregation were encountered; their minimisation and the subsequent estimation of the kinetic parameters for the interaction of lysostaphin with the ***substrates*** is described, as well as a comparison of ***substrates*** incorporating two ***FRET pairs***: Abz-EDDnp and ***DABCYL-EDANS***

. In addition to this, the points of cleavage caused by lysostaphin in Abz-pentaglycine-EDDnp have been determined by HPLC and mass spectrometry analysis to be between glycines 2 and 3(approximately 60%) and glycines 3 and 4 (approximately 40%).

L11 ANSWER 2 OF 4 CAPLUS COPYRIGHT 2006 ACS on STN
AB Lysostaphin (EC. 3.4.24.75) is a protein secreted by Staphylococcus simulans biovar staphylolyticus and has been shown to be active against methicillin resistant S. aureus (MRSA). The design and synthesis of three internally quenched ***substrates*** for lysostaphin based on the peptidoglycan crossbridges of S. aureus, and their use in fluorescence resonance energy transfer (FRET) assays is reported. These ***substrates*** enabled the gathering of information about the endopeptidase activity of lysostaphin and the effect that mutations have on its enzymic ability. Significant problems with the inner filter effect and ***substrate*** aggregation were encountered; their minimization and the subsequent estimation of the kinetic parameters for the interaction of lysostaphin with the ***substrates*** is described, as well as a comparison of ***substrates*** incorporating two ***FRET pairs***: Abz-EDDnp and ***DABCYL-EDANS***. In addition to this, the points of cleavage caused by lysostaphin in Abz-pentaglycine-EDDnp have been determined by HPLC and mass spectrometry anal. to be between glycines 2 and 3 (.apprx.60%) and glycines 3 and 4 (.apprx.40%).

L11 ANSWER 3 OF 4 CAPLUS COPYRIGHT 2006 ACS on STN
AB Compns. and methods are provided for identifying conformational changes in polypeptides related to the activity or biol. state of the polypeptide. Semisynthetic polypeptides are prepared comprising at least two proximity-sensor peptides, the resultant composition capable of detectably indicating the activity of biol. state of the polypeptide. Such compns. may be used to identify modulators of the polypeptides as well as modulators of mols. which interact with the polypeptide, such as protein kinases which act on protein kinase targets. More specifically, the invention provides a biosensor for c-Abl phosphorylation of the Crk-II adapter protein. The structure of a dual-labeled, semisynthetic, recombinantly prepared composition comprising the protein kinase adapter protein Crk-II which is capable of reporting phosphorylation by c-Abl is disclosed. Generation of Rh-(Crk-II)-Fl (Rh = tetramethylrhodamine; Fl = fluorescein) by solid-phase protein ligation and phosphorylation of Rh-(Crk-II)-Fl by full-length c-Abl is reported. It was shown that Rh-(Crk-II)-Fl is a fluorescence biosensor for c-Abl phosphorylation of Crk-II utilizing FRET for c-Abl determination One potential use of this biosensor is in the rapid screening of c-Abl kinase inhibitors.

L11 ANSWER 4 OF 4 CAPLUS COPYRIGHT 2006 ACS on STN
AB A method of assaying for enzymes that remove purine or pyrimidine bases from nucleic acids in a sequence-specific manner without strand scission is described. The method uses a mol. beacon as a ***substrate***. A stem-loop nucleic acid structure brings together a ***FRET pair*** of reporter dyes: a fluorophore and a quencher. As the bases are removed from the stem by the enzyme, the stem loses stability and the fluorophore is able to fluoresce. Use of a fluorescein/***DABCYL*** pair to assay the rRNA N-glycosidase activity of viscumin is demonstrated.

=> s l1 and l5
L12 0 L1 AND L5

=> s l1 and l6
L13 0 L1 AND L6

```
=> s l1 and l7
L14           1 L1 AND L7

=> d all

L14  ANSWER 1 OF 1  CAPLUS  COPYRIGHT 2006 ACS on STN
AN   2006:170631  CAPLUS
DN   144:249410
ED   Entered STN:  24 Feb 2006
TI   FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing
     semisynthetic dual-labeled proximity sensor peptide-containing Crk-II
     construct, and potential screening use
IN   Muir, Tom; Cotton, Graham
PA   USA
SO   U.S. Pat. Appl. Publ., 19 pp., Cont. of U.S. Ser. No. 483,543, abandoned.
     CODEN: USXXCO
DT   Patent
LA   English
INCL 435007100
CC   7-1 (Enzymes)
     Section cross-reference(s): 1, 34
FAN.CNT 1
     PATENT NO.          KIND   DATE        APPLICATION NO.        DATE
     ---------------     ----   --------    --------------------   --------
PI   US 2006040319        A1    20060223    US 2004-784721         20040223
PRAI US 2000-483543       B1    20000114
CLASS
 PATENT NO.       CLASS  PATENT FAMILY CLASSIFICATION CODES
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 US 2006040319    INCL   435007100
                  IPCI   G01N0033-53 [I,A]
                  IPCR   G01N0033-53 [I,A]; G01N0033-53 [I,C]
                  NCL    435/007.100
                  ECLA   C12Q001/48B; G01N033/542
AB   Compns. and methods are provided for identifying conformational changes in
     polypeptides related to the activity or biol. state of the polypeptide.
     Semisynthetic polypeptides are prepared comprising at least two
     proximity-sensor peptides, the resultant composition capable of detectably
     indicating the activity of biol. state of the polypeptide.  Such compns.
     may be used to identify modulators of the polypeptides as well as
     modulators of mols. which interact with the polypeptide, such as protein
     kinases which act on protein kinase targets.  More specifically, the
     invention provides a biosensor for c-Abl phosphorylation of the Crk-II
     adapter protein.  The structure of a dual-labeled, semisynthetic,
     recombinantly prepared composition comprising the protein kinase adapter
protein
     Crk-II which is capable of reporting phosphorylation by c-Abl is
     disclosed.  Generation of Rh-(Crk-II)-Fl (Rh = tetramethylrhodamine; Fl =
     fluorescein) by solid-phase protein ligation and phosphorylation of
     Rh-(Crk-II)-Fl by full-length c-Abl is reported.  It was shown that
     Rh-(Crk-II)-Fl is a fluorescence biosensor for c-Abl phosphorylation of
     Crk-II utilizing FRET for c-Abl determination  One potential use of this
biosensor
     is in the rapid screening of c-Abl kinase inhibitors.
ST   fluorescence biosensor cAbl kinase detn CrkII phosphorylation FRET;
     proximity sensor peptide fluorescence biosensor protein kinase detn
     screening; FRET pair tetramethylrhodamine fluorescein
     proximity sensor peptide fluorescence biosensor
IT   Proteins
     RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); BSU
     (Biological study, unclassified); PRP (Properties); SPN (Synthetic
     preparation); ANST (Analytical study); BIOL (Biological study); PREP
     (Preparation); USES (Uses)
        (CRKII, fusion products; FRET assay for c-Abl phosphorylation of Crk-II
```

adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Mus musculus
(Crk-II of; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Dephosphorylation, biological
Drug screening
Fluorescent indicators
Phosphorylation, biological
Protein engineering
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Enzymes, analysis
RL: ANT (Analyte); ANST (Analytical study)
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein motifs
(SH2 domain; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein motifs
(SH3 domain; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Conformational transition
(activity-related, detection of; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Phosphorylation
(enzymic; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Fluorescence resonance energy transfer
(fluorescent label pair for; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein sequences
(of Crk-II construct; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Post-translational processing
(of peptide ***substrate***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Biosensors
(optical; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Solid phase synthesis
(peptide, solid-phase protein ligation; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Amino acids, uses
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(proximity sensor peptide containing fluorescent amino acid derivative; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Oligopeptides
RL: ARG (Analytical reagent use); SPN (Synthetic preparation); ANST (Analytical study); PREP (Preparation); USES (Uses)
(proximity sensor; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Proteins
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(recombinant, ***substrates***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Peptides, uses
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(***substrate***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT ***Phycoerythrins***
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(β-, fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 146368-14-1, ***Cy5***
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(***Cy5***, fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 9031-44-1, Kinase (phosphorylating) 98037-52-6, Abelson protein tyrosine kinase 138238-67-2, c-Abl kinase
RL: ANT (Analyte); ANST (Analytical study)
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 876799-69-8P 876799-70-1P
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); SPN (Synthetic preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(amino acid sequence; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 91-64-5, Coumarin 129-00-0, Pyrene, uses 2321-07-5, Fluorescein 6268-49-1, DABCYL 36930-63-9, IAEDANS 50402-56-7, EDANS 70281-37-7, Tetramethylrhodamine 138026-71-8, BODIPY 165599-63-3, BODIPY FL
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 876656-99-4D, conjugates with fluorescein
RL: RCT (Reactant); RACT (Reactant or reagent)
(preparation of dual-labeled Crk-II; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 876808-11-6
RL: PRP (Properties)
(unclaimed protein sequence; fRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 168202-46-8 302566-59-2 876760-73-5 876760-74-6 876808-08-1 876808-09-2 876808-10-5

RL: PRP (Properties)
(unclaimed sequence; fRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

=> s l1 and l9
L15 1 L1 AND L9

=> d all

L15 ANSWER 1 OF 1 CAPLUS COPYRIGHT 2006 ACS on STN
AN 2006:170631 CAPLUS
DN 144:249410
ED Entered STN: 24 Feb 2006
TI FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use
IN Muir, Tom; Cotton, Graham
PA USA
SO U.S. Pat. Appl. Publ., 19 pp., Cont. of U.S. Ser. No. 483,543, abandoned.
CODEN: USXXCO
DT Patent
LA English
INCL 435007100
CC 7-1 (Enzymes)
Section cross-reference(s): 1, 34
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI US 2006040319	A1	20060223	US 2004-784721	20040223
PRAI US 2000-483543	B1	20000114		

CLASS

PATENT NO.	CLASS	PATENT FAMILY CLASSIFICATION CODES
US 2006040319	INCL	435007100
	IPCI	G01N0033-53 [I,A]
	IPCR	G01N0033-53 [I,A]; G01N0033-53 [I,C]
	NCL	435/007.100
	ECLA	C12Q001/48B; G01N033/542

AB Compns. and methods are provided for identifying conformational changes in polypeptides related to the activity or biol. state of the polypeptide. Semisynthetic polypeptides are prepared comprising at least two proximity-sensor peptides, the resultant composition capable of detectably indicating the activity of biol. state of the polypeptide. Such compns. may be used to identify modulators of the polypeptides as well as modulators of mols. which interact with the polypeptide, such as protein kinases which act on protein kinase targets. More specifically, the invention provides a biosensor for c-Abl phosphorylation of the Crk-II adapter protein. The structure of a dual-labeled, semisynthetic, recombinantly prepared composition comprising the protein kinase adapter protein Crk-II which is capable of reporting phosphorylation by c-Abl is disclosed. Generation of Rh-(Crk-II)-Fl (Rh = tetramethylrhodamine; Fl = ***fluorescein***) by solid-phase protein ligation and phosphorylation of Rh-(Crk-II)-Fl by full-length c-Abl is reported. It was shown that Rh-(Crk-II)-Fl is a fluorescence biosensor for c-Abl phosphorylation of Crk-II utilizing FRET for c-Abl determination One potential use of this biosensor is in the rapid screening of c-Abl kinase inhibitors.
ST fluorescence biosensor cAbl kinase detn CrkII phosphorylation FRET; proximity sensor peptide fluorescence biosensor protein kinase detn screening; ***FRET pair*** tetramethylrhodamine ***fluorescein*** proximity sensor peptide fluorescence biosensor

IT Proteins
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); SPN (Synthetic preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(CRKII, fusion products; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Mus musculus
(Crk-II of; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Dephosphorylation, biological
Drug screening
Fluorescent indicators
Phosphorylation, biological
Protein engineering
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Enzymes, analysis
RL: ANT (Analyte); ANST (Analytical study)
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein motifs
(SH2 domain; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein motifs
(SH3 domain; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Conformational transition
(activity-related, detection of; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Phosphorylation
(enzymic; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Fluorescence resonance energy transfer
(fluorescent label pair for; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Protein sequences
(of Crk-II construct; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Post-translational processing
(of peptide ***substrate***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Biosensors
(optical; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Solid phase synthesis
(peptide, solid-phase protein ligation; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Amino acids, uses
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(proximity sensor peptide containing fluorescent amino acid derivative; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Oligopeptides
RL: ARG (Analytical reagent use); SPN (Synthetic preparation); ANST (Analytical study); PREP (Preparation); USES (Uses)
(proximity sensor; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Proteins
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(recombinant, ***substrates***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Peptides, uses
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(***substrate***; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT Phycoerythrins
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(β-, fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 146368-14-1, Cy5
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(Cy5, fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 9031-44-1, Kinase (phosphorylating) 98037-52-6, Abelson protein tyrosine kinase 138238-67-2, c-Abl kinase
RL: ANT (Analyte); ANST (Analytical study)
(FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 876799-69-8P 876799-70-1P
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); SPN (Synthetic preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(amino acid sequence; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 91-64-5, Coumarin 129-00-0, Pyrene, uses 2321-07-5, ***Fluorescein*** 6268-49-1, DABCYL 36930-63-9, ***IAEDANS*** 50402-56-7, EDANS 70281-37-7, Tetramethylrhodamine 138026-71-8, BODIPY 165599-63-3, BODIPY FL
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 876656-99-4D, conjugates with ***fluorescein***
RL: RCT (Reactant); RACT (Reactant or reagent)
(preparation of dual-labeled Crk-II; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 876808-11-6
RL: PRP (Properties)
(unclaimed protein sequence; fRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)
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876808-09-2 876808-10-5
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=> s l1 and l8
L16 2 L1 AND L8

=> d all

L16 ANSWER 1 OF 2 CAPLUS COPYRIGHT 2006 ACS on STN
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PRAI US 2000-483543	B1	20000114		

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	ECLA	C12Q001/48B; G01N033/542

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Crk-II utilizing FRET for c-Abl determination One potential use of this biosensor is in the rapid screening of c-Abl kinase inhibitors.

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(β-, fluorescent label for FRET; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

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IT 876799-69-8P 876799-70-1P
RL: ARG (Analytical reagent use); BPN (Biosynthetic preparation); BSU (Biological study, unclassified); PRP (Properties); SPN (Synthetic preparation); ANST (Analytical study); BIOL (Biological study); PREP (Preparation); USES (Uses)
(amino acid sequence; FRET assay for c-Abl phosphorylation of Crk-II adapter protein utilizing semisynthetic dual-labeled proximity sensor peptide-containing Crk-II construct, and potential screening use)

IT 91-64-5, ***Coumarin*** 129-00-0, ***Pyrene***, uses
2321-07-5, Fluorescein 6268-49-1, DABCYL 36930-63-9, IAEDANS
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RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
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